Copy for the Elected Office (EO/US)

PCT/US99/27919

PATENT COOPERATION TREATY

From the INTERNATIONAL BUREAU

PCT

NOTIFICATION OF THE RECORDING OF A CHANGE

(PCT Rule 92bis.1 and Administrative Instructions, Section 422)

To:

GOLDHUSH, Douglas, H.
Arent Fox Kintner Plotkin & Kahn, PLLC
1050 Connecticut Avenue, NW
Suite 600
Washington, DC 20036-5339
ETATS-UNIS D'AMERIQUE

Date of mailing (day/month/year)
25 September 2000 (25.09.00)

Applicant's or agent's file reference
F8172-9032

IMPORTANT NOTIFICATION

International application No.
PCT/US99/27919

International filing date (day/month/year)
08 December 1999 (08.12.99)

1. The following indications appeared on record concerning:

☐ the applicant ☐ the inventor ☒ the agent ☐ the common representative

Name and Address

GOLDHUSH, Douglas, H.
Arent Fox Kitner Plotkin & Kahn, PLLC
1050 Connecticut Avenue, NW
Suite 600
Washington, DC 20005-5701
United States of America

State of Nationality:

State of Residence:

Telephone No.: 202 638 5000

Facsimile No.: 202 638 4810

Teleprinter No.:

2. The International Bureau hereby notifies the applicant that the following change has been recorded concerning:

☐ the person ☐ the name ☒ the address ☐ the nationality ☐ the residence

Name and Address

GOLDHUSH, Douglas, H.
Arent Fox Kintner Plotkin & Kahn, PLLC
1050 Connecticut Avenue, NW
Suite 600
Washington, DC 20036-5339
United States of America

State of Nationality:

State of Residence:

Telephone No.: 202 857 6000

Facsimile No.: 202 638 4810

Teleprinter No.:

3. Further observations, if necessary:

4. A copy of this notification has been sent to:

☒ the receiving Office
☐ the International Searching Authority
☒ the International Preliminary Examining Authority
☐ the designated Offices concerned
☒ the elected Offices concerned
☐ other:

The International Bureau of WIPO
34, chemin des Colombettes
1211 Geneva 20, Switzerland

Facsimile No.: (41-22) 740.14.35

Authorized officer

Jocelyne Rey-Millet

Telephone No.: (41-22) 338.83.38

PATENT COOPERATION TREATY

From the INTERNATIONAL BUREAU

PCT

NOTIFICATION OF ELECTION

(PCT Rule 61.2)

To:

Assistant Commissioner for Patents
United States Patent and Trademark Office
Box PCT
Washington, D.C.20231
ETATS-UNIS D'AMERIQUE

in its capacity as elected Office

Date of mailing (day/month/year) 24 August 2000 (24.08.00)	
International application No. PCT/US99/27919	**Applicant's or agent's file reference** F8172-9032
International filing date (day/month/year) 08 December 1999 (08.12.99)	**Priority date** (day/month/year) 08 December 1998 (08.12.98)
Applicant HAGBERG, James, M. et al	

1. The designated Office is hereby notified of its election made:

[X] in the demand filed with the International Preliminary Examining Authority on:

07 July 2000 (07.07.00)

[ ] in a notice effecting later election filed with the International Bureau on:

2. The election [X] was

[ ] was not

made before the expiration of 19 months from the priority date or, where Rule 32 applies, within the time limit under Rule 32.2(b).

The International Bureau of WIPO **34, chemin des Colombettes** **1211 Geneva 20, Switzerland**	Authorized officer Manu Berrod
Facsimile No.: (41-22) 740.14.35	Telephone No.: (41-22) 338.83.38

PCT WORLD INTELLECTUAL PROPERTY ORGANIZATION
International Bureau

INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification 7 : C12Q 1/68, C07H 21/04

A1

(11) International Publication Number: **WO 00/34520**

(43) International Publication Date: 15 June 2000 (15.06.00)

(21) International Application Number: PCT/US99/27919

(22) International Filing Date: 8 December 1999 (08.12.99)

(30) Priority Data:

60/111,494	8 December 1998 (08.12.98)	US
60/112,604	17 December 1998 (17.12.98)	US

(71) Applicants *(for all designated States except US)*: UNIVERSITY OF MARYLAND, COLLEGE PARK [US/US]; Office of Technology Liaison, 4312 Knox Road, College Park, MD 20742 (US). UNIVERSITY OF MARYLAND, BALTIMORE [US/US]; Office of Research & Development, 515 West Lombard Street, Baltimore, MD 21201 (US). UNIVERSITY OF PITTSBURGH [US/US]; Office of Technology Management, Rm 9110, William Pitt Union, Pittsburgh, PA 15260 (US).

(72) Inventors; and

(75) Inventors/Applicants *(for US only)*: HAGBERG, James, M. [US/US]; 10941 Hilltop Lane, Columbia, MD 21044 (US). FERRELL, Robert, E. [US/US]; 206 Maple Avenue, Pittsburgh, PA 15218 (US). SHULDINER, Alan [US/US]; 10600 Harpoon Hill, Columbia, MD 21044 (US).

(74) Agents: GOLDHUSH, Douglas, H. et al.; Nikaido, Marmelstein, Murray & Oram LLP, Metropolitan Square, 655 15th Street N.W., G Street Lobby, Suite 330, Washington, DC 20005–5701 (US).

(81) Designated States: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CR, CU, CZ, DE, DK, DM, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, ARIPO patent (GH, GM, KE, LS, MW, SD, SL, SZ, TZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG).

Published

With international search report.

Before the expiration of the time limit for amending the claims and to be republished in the event of the receipt of amendments.

(54) Title: GENETIC MARKERS WHICH IDENTIFY INDIVIDUALS WHO IMPROVE THEIR CHOLESTEROL LEVELS AND DIABETES STATUS WITH EXERCISE

(57) Abstract

A method of improving cholesterol levels or diabetes status in subjects with hypercholesteremia or diabetes or at risk of developing such disorders includes identifying subjects having an allele and/or a genotype at a gene locus which positively correlates with greater success in improving cholesterol levels or diabetes status in hypercholesteremic or diabetic individuals, respectively, as compared with other alleles and/or genotypes at the same gene locus, and engaging the subject in exercise training for a period of time sufficient to improve the subject's cholesterol levels or diabetes status. Genotypic distinctions may be found, for example, in the glucose transport 4 gene, the myostatin exon 2 gene and the insulin receptor substrate–1 (IRS–1) gene.

FOR THE PURPOSES OF INFORMATION ONLY

Codes used to identify States party to the PCT on the front pages of pamphlets publishing international applications under the PCT.

AL	Albania	**ES**	Spain	**LS**	Lesotho	**SI**	Slovenia
AM	Armenia	**FI**	Finland	**LT**	Lithuania	**SK**	Slovakia
AT	Austria	**FR**	France	**LU**	Luxembourg	**SN**	Senegal
AU	Australia	**GA**	Gabon	**LV**	Latvia	**SZ**	Swaziland
AZ	Azerbaijan	**GB**	United Kingdom	**MC**	Monaco	**TD**	Chad
BA	Bosnia and Herzegovina	**GE**	Georgia	**MD**	Republic of Moldova	**TG**	Togo
BB	Barbados	**GH**	Ghana	**MG**	Madagascar	**TJ**	Tajikistan
BE	Belgium	**GN**	Guinea	**MK**	The former Yugoslav Republic of Macedonia	**TM**	Turkmenistan
BF	Burkina Faso	**GR**	Greece			**TR**	Turkey
BG	Bulgaria	**HU**	Hungary	**ML**	Mali	**TT**	Trinidad and Tobago
BJ	Benin	**IE**	Ireland	**MN**	Mongolia	**UA**	Ukraine
BR	Brazil	**IL**	Israel	**MR**	Mauritania	**UG**	Uganda
BY	Belarus	**IS**	Iceland	**MW**	Malawi	**US**	United States of America
CA	Canada	**IT**	Italy	**MX**	Mexico	**UZ**	Uzbekistan
CF	Central African Republic	**JP**	Japan	**NE**	Niger	**VN**	Viet Nam
CG	Congo	**KE**	Kenya	**NL**	Netherlands	**YU**	Yugoslavia
CH	Switzerland	**KG**	Kyrgyzstan	**NO**	Norway	**ZW**	Zimbabwe
CI	Côte d'Ivoire	**KP**	Democratic People's Republic of Korea	**NZ**	New Zealand		
CM	Cameroon			**PL**	Poland		
CN	China	**KR**	Republic of Korea	**PT**	Portugal		
CU	Cuba	**KZ**	Kazakstan	**RO**	Romania		
CZ	Czech Republic	**LC**	Saint Lucia	**RU**	Russian Federation		
DE	Germany	**LI**	Liechtenstein	**SD**	Sudan		
DK	Denmark	**LK**	Sri Lanka	**SE**	Sweden		
EE	Estonia	**LR**	Liberia	**SG**	Singapore		

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US99/27919

A. CLASSIFICATION OF SUBJECT MATTER

IPC(7) : C12Q 1/68; C07H 21/04
US CL : 435/6; 536/23.5

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
U.S. : 435/6; 536/23.5

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)
Please See Continuation Sheet

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	BJORBAEK et al. Genetic Variants in Promoters and Coding Regions of the Muscle Glycogen Synthase and the Insulin-Responsive GLUT4 Genes in NIDDM. Diabetes. August 1994, Vol. 43, pages 976-983.	1
A	OSSEI-GERNING et al. Insulin receptor substrate-1 gene polymorphism and cardiovascular risk in non-insulin dependent diabetes mellitus and patients undergoing coronary angiography. Clin. Lab. Haem. 1997, Vol. 19, pages 123-129.	3
A	GROBET et al. Molecular definition of an allelic series of mutations disrupting the myostatin function and causing double-muscling in cattle. Mammalian Genome. 1998, Vol. 9, pages 210-213.	2, 4

☐ Further documents are listed in the continuation of Box C. ☐ See patent family annex.

* Special categories of cited documents:

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier application or patent published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
08 March 2000 (08.03.2000)	05 APR 2000
Name and mailing address of the ISA/US Commissioner of Patents and Trademarks Box PCT Washington, D.C. 20231 Facsimile No. (703) 305-3230	Authorized officer Juliet C. Einsmann Telephone No. (703) 308-0196

Form PCT/ISA/210 (second sheet) (July 1998)

INTERNATIONAL SEARCH REPORT

International application No.

PCT/US99/27919

Continuation of B. FIELDS SEARCHED Item 3: MEDLINE, BIOSIS, EMBASE, CAPLUS, SCISEARCH
search terms: cholesterol, exercise, diabetes, hypercholestermia, glucose transport, myostatin, insulin receptor substrate

PATENT COOPERATION TREATY

From the
INTERNATIONAL PRELIMINARY EXAMINING AUTHORITY

To:
DOUGLAS H. GOLDHUSH
ARENT FOX KINTNER PLOTKIN & KAHN PLLC
1050 CONNECTICUT AVENUE NW
SUITE 600
WASHINGTON D.C. 20036-5339

PCT

NOTIFICATION OF TRANSMITTAL OF INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Rule 71.1)

Date of Mailing *(day/month/year)*	16FEB2001
Applicant's or agent's file reference 108172-09032	**IMPORTANT NOTIFICATION**

International application No.	International filing date *(day/month/year)*	Priority date *(day/month/year)*
PCT/US99/27919	08 December 1999 (08.12.1999)	08 December 1998 (08.12.1998)

Applicant

UNIVERSITY OF MARYLAND, COLLEGE PARK

1. The applicant is hereby notified that this International Preliminary Examining Authority transmits herewith the international preliminary examination report and its annexes, if any, established on the international application.

2. A copy of the report and its annexes, if any, is being transmitted to the International Bureau for communication to all the elected Offices.

3. Where required by any of the elected Offices, the International Bureau will prepare an English translation of the report (but not of any annexes) and will transmit such translation to those Offices.

4. **REMINDER**

The applicant must enter the national phase before each elected Office by performing certain acts (filing translations and paying national fees) within 30 months from the priority date (or later in some Offices)(Article 39(1))(see also the reminder sent by the International Bureau with Form PCT/IB/301).

Where a translation of the international application must be furnished to an elected Office, that translation must contain a translation of any annexes to the international preliminary examination report. It is the applicant's responsibility to prepare and furnish such translation directly to each elected Office concerned.

For further details on the applicable time limits and requirements of the elected Offices, see Volume II of the PCT Applicant's Guide.

Name and mailing address of the IPEA/US Commissioner of Patents and Trademarks Box PCT Washington, D.C. 20231 Facsimile No. (703)305-3230	Authorized officer Juliet C. Einsmann Telephone No. (703) 308-0196

TERRY J. DEY
PARALEGAL SPECIALIST
TECHNOLOGY CENTER 1600

Form PCT/IPEA/416 (July 1992)

PATENT COOPERATION TREATY

PCT

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Article 36 and Rule 70)

Applicant's or agent's file reference 108172-09032	**FOR FURTHER ACTION**	See Notification of Transmittal of International Preliminary Examination Report (Form PCT/IPEA/416)
International application No. PCT/US99/27919	International filing date *(day/month/year)* 08 December 1999 (08.12.1999)	Priority date *(day/month/year)* 08 December 1998 (08.12.1998)
International Patent Classification (IPC) or national classification and IPC IPC(7): C12Q 1/68; C07H 21/04 and US Cl.: 435/6; 536/23.5		
Applicant UNIVERSITY OF MARYLAND, COLLEGE PARK		

1. This international preliminary examination report has been prepared by this International Preliminary Examining Authority and is transmitted to the applicant according to Article 36.

2. This REPORT consists of a total of 3 sheets, including this cover sheet.

   ☐ This report is also accompanied by ANNEXES, i.e., sheets of the description, claims and/or drawings which have been amended and are the basis for this report and/or sheets containing rectifications made before this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions under the PCT).

   These annexes consist of a total of 0 sheets.

3. This report contains indications relating to the following items:

   - I ☒ Basis of the report
   - II ☐ Priority
   - III ☐ Non-establishment of report with regard to novelty, inventive step and industrial applicability
   - IV ☐ Lack of unity of invention
   - V ☒ Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
   - VI ☐ Certain documents cited
   - VII ☐ Certain defects in the international application
   - VIII ☐ Certain observations on the international application

Date of submission of the demand	Date of completion of this report
07 July 2000 (07.07.2000)	24 November 2000 (24.11.2000)
Name and mailing address of the IPEA/US Commissioner of Patents and Trademarks Box PCT Washington, D.C. 20231 Facsimile No. (703)305-3230	Authorized officer Juliet C. Einsmann Telephone No. (703) 308-01

TERRY J. DEY
PARALEGAL SPECIALIST
TECHNOLOGY CENTER 1600

Form PCT/IPEA/409 (cover sheet)(July 1998)

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

International application No.
PCT/US99/27919

I. Basis of the report

1. With regard to the **elements** of the international application:*

☒ the international application as originally filed.

☒ the description:
pages 1-10 as originally filed
pages NONE , filed with the demand
pages NONE , filed with the letter of _____.

☒ the claims:
pages 11 and 12 , as originally filed
pages NONE , as amended (together with any statement) under Article 19
pages NONE , filed with the demand
pages NONE , filed with the letter of _____.

☒ the drawings:
pages NONE , as originally filed
pages NONE , filed with the demand
pages NONE , filed with the letter of _____.

☐ the sequence listing part of the description:
pages NONE , as originally filed
pages NONE , filed with the demand
pages NONE , filed with the letter of _____.

2. With regard to the **language**, all the elements marked above were available or furnished to this Authority in the language in which the international application was filed, unless otherwise indicated under this item.
These elements were available or furnished to this Authority in the following language _____ which is:

☐ the language of a translation furnished for the purposes of international search (under Rule23.1(b)).

☐ the language of publication of the international application (under Rule 48.3(b)).

☐ the language of the translation furnished for the purposes of international preliminary examination(under Rules 55.2 and/or 55.3).

3. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, the international preliminary examination was carried out on the basis of the sequence listing:

☐ contained in the international application in printed form.

☐ filed together with the international application in computer readable form.

☐ furnished subsequently to this Authority in written form.

☐ furnished subsequently to this Authority in computer readable form.

☐ The statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.

☐ The statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished.

4. ☐ The amendments have resulted in the cancellation of:

☐ the description, pages NONE

☐ the claims, Nos. NONE

☐ the drawings, sheets/~~fig~~ NONE

5. ☐ This report has been established as if (some of) the amendments had not been made, since they have been considered to go beyond the disclosure as filed, as indicated in the Supplemental Box (Rule 70.2(c)).**

** Replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report since they do not contain amendments (Rules 70.16 and 70.17).*
*** Any replacement sheet containing such amendments must be referred to under item 1 and annexed to this report.*

INTERNATIONAL PRELIM[illegible]ARY EXAMINATION REPORT

Internat[illegible]application No.
PCT/US99/27919

V. Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. STATEMENT

Novelty (N)	Claims 1-4	YES
	Claims NONE	NO
Inventive Step (IS)	Claims 1-4	YES
	Claims NONE	NO
Industrial Applicability (IA)	Claims 1-4	YES
	Claims NONE	NO

2. CITATIONS AND EXPLANATIONS (Rule 70.7)

Claims 1-4 meet the criteria set out in PCT Articles 33(2)-(3) since the prior art does not teach or fairly suggest the claimed invention. The cited references teach mutations in the glucose transport 4 gene (Bjorbaek et al.), the myostatin exon 2 gene (Grobet et al.), and the insulin receptor substrate-1 gene (Ossei-Gerning et al.). The prior art fails to teach, however, an association between these mutations and high cholesterol or that people with specific genotypes in these genes will be more responsive to specific exercise levels for improving choleterol levels.

Claims 1-4 meet the criteria set out in PCT Article 33(4) for industrial applicability.

---------- NEW CITATIONS ----------

NONE

PATENT COOPERATION TREATY

From the
INTERNATIONAL PRELIMINARY EXAMINING AUTHORITY

To:
DOUGLAS H. GOLDHUSH
NIKAIDO, MARMELSTEIN, MURRAY & ORAM LLP
METROPOLITAN SQUARE; G STREET LOBBY
655 FIFTEENTH STREET, N.W., SUITE 330
WASHINGTON, DC 20005 5701

PCT

NOTIFICATION OF RECEIPT
OF DEMAND BY COMPETENT INTERNATIONAL
PRELIMINARY EXAMINING AUTHORITY

(PCT Rules 59.3(e) and 61.1(b), first sentence
and Administrative Instructions, Section 601(a))

Date of mailing *(day/month/year)*	07 AUG 2000

Applicant's or agent's file reference	IMPORTANT NOTIFICATION
F8172-9032	

International application No.	International filing date *(day/month/year)*	Priority date *(day/month/year)*
PCT/US99/27919	08 DEC 99	08 DEC 98

Applicant
UNIVERSITY OF MARYLAND, COLLEGE PARK

1. The applicant is hereby **notified** that this International Preliminary Examining Authority considers the following date as the date of receipt of the demand for international preliminary examination of the international application:

   07 JULY 2000 (07.07.00)

2. That date of receipt is:

   [X] the actual date of receipt of the demand by this Authority (Rule 61.1(b)).

   [ ] the actual date of receipt of the demand on behalf of this Authority (Rule 59.3(e)).

   [ ] the date on which this Authority has, in response to the invitation to correct defects in the demand (Form PCT/IPEA/404), received the required corrections.

3. [ ] **ATTENTION:** That date of receipt is **AFTER** the expiration of 19 months from the priority date. Consequently, the election(s) made in the demand does (do) not have the effect of postponing the entry into the national phase until 30 months from the priority date (or later in some Offices) (Article 39(1)). Therefore, the acts for entry into the national phase must be performed within 20 months from the priority date (or later in some Offices) (Article 22). For details, see the *PCT Applicant's Guide*, Volume II.

   [ ] *(If applicable)* This notification confirms the information given by telephone, facsimile transmission or in person on:

   _____

4. Only where paragraph 3 applies, a copy of this notification has been sent to the International Bureau.

Name and mailing address of the IPEA/ Assistant Commissioner for Patent Box PCT Washington, D.C. 20231 Attn:RO/US Facsimile No. 703-305-3230	Authorized officer: Felicia Lawrence PCT Operations - IAP Team 1 (703) 305-3675 (703) 305-3230 (FAX Telephone No.

Form PCT/IPEA/402 (July 1998)

PATENT COOPERATION TREATY

From the
INTERNATIONAL PRELIMINARY EXAMINING AUTHORITY

To:
DOUGLAS H. GOLDHUSH
NIKAIDO, MARMELSTEIN, MURRAY & ORAM LLP
METROPOLITAN SQUARE; G STREET LOBBY
655 FIFTEENTH STREET, N.W., SUITE 330
WASHINGTON, DC 20005 5701

PCT

INVITATION TO CORRECT
DEFECTS IN THE DEMAND

(PCT Rule 60.1)

	Date of mailing *(day/month/year)* 07 AUG 2000
Applicant's or agent's file reference F8172-9032	**REPLY DUE** within **ONE MONTH** from the above date of mailing. See also below.
International application No. PCT/US99/27919	International filing date *(day month/year)* 08 DEC 99
Applicant UNIVERSITY OF MARYLAND, COLLEGE PARK	

The applicant is hereby **invited** within the time limit indicated above **to correct the following defects** which this International Preliminary Examining Authority has found in the demand for international preliminary examination:

1. ☐ It does not contain the election of at least one Contracting State bound by Chapter II (Rules 53.2(a)(iv) and 53.7).
2. ☐ It does not permit identification of the international application to which it relates (Rule 60.1(b)).
3. ☐ It does not contain the required petition (Rules 53.2(a)(i) and 53.3).
4. ☒ It does not contain the required indications concerning the agent as specified in the Annex (Rules 53.2(a)(ii) and 53.5).
5. ☐ It does not contain the required indications concerning the international application as specified in the Annex (Rules 53.2(a)(iii) and 53.6).
6. ☐ It is not submitted in the required language which is: _____ (Rule 55.1).
7. ☐ It is not made on the printed form (Rule 53.1(a)).
8. ☐ It is presented as a computer print-out the particulars of which do not comply with the Administrative Instructions (Rule 53.1(a)).
9. ☐ It does not contain the required indications concerning the applicant as specified in the Annex (Rules 53.2(a)(ii) and 53.4).
10. ☐ It does not contain the required signature as specified in the Annex (Rules 53.2(b) and 53.8).

Effect of the date of receipt of the corrections on the date of receipt of the demand:

(i) If the defects noted under items 1 and 2 are corrected within the time limit indicated above, the demand shall be considered as if it had been received on the date when the corrections are received (Rule 60.1(b)). If that date is later than the expiration of 19 months from the priority date, entry into the national phase before the elected Offices will **NOT** be postponed until the expiration of 30 months from the priority date.

(ii) If the defects noted under items 3 to 10 are corrected within the time limit indicated above, the demand shall be considered as if it had been received on the actual filing date (Rule 60.1(b)).

Effect of failure to correct the defects within the time limit indicated above:

(i) In the case of defects noted under items 1 to 8, this Authority will declare that the demand is considered as if it had not been submitted.

(ii) In the case of defects noted under items 9 and 10, this Authority will declare that the election(s) of the State(s) concerned is(are) considered as if it(they) had not been made.

A copy of this invitation has been sent to the International Bureau.

Name and mailing address of the IPEA/ Assistant Commissioner for Patent Box PCT Washington, D.C. 20231 Attn:RO/US Facsimile No. 703-305-3230	Authorized officer: Felicia Lawrence PCT Operations - IAPD Team 1 Telephone No. (703) 305-3675 (703) 305-3230 (FAX)

Form PCT/IPEA/404 (July 1998)

ANNEX TO
FORM PCT/IPEA/404

International application No.
PCT/US99/27919

Continuation of item 4: As to indications concerning **the agent** (Rule 4.4), the demand:

a. ☐ does not properly indicate the agent's name *(specify)*:

b. ☐ does not indicate the agent's address.

c. ☒ does not properly indicate the agent's address *(specify)*:
THE ADDRESS ON THE DEMAND IS DIFFER FROM WHATS ON THE REQUEST. (PLEASE CLARIFY)

Continuation of item 5: As to indications concerning **the international application**, the demand does not indicate:

a. ☐ the international filing date.

b. ☐ the international application number.

c. ☐ the name of the receiving Office, where the international application number was not known to the applicant at the time the demand was filed.

d. ☐ the title of the invention.

Continuation of item 9: As to indications concerning **the applicant** (Rules 4.4 and 4.5), the demand:

a. ☐ does not indicate all the applicants for the elected States.

b. ☐ does not properly indicate the applicant's name *(specify)*:

c. ☐ does not indicate the applicant's address.

d. ☐ does not properly indicate the applicant's address *(specify)*:

e. ☐ does not indicate the applicant's nationality.

f. ☐ does not indicate the applicant's residence.

Continuation of item 10: As to requirements concerning **signature** (Rules 4.15 and 90.4), the demand:

a. ☐ is not signed.

b. ☐ is not signed by all the applicants for the elected States.

c. ☐ is not accompanied by the statement referred to in the check list in Box No. VI of the demand explaining the lack of the signature of an applicant for the election of the United States of America.

d. ☐ is signed by what appears to be an agent/common representative but

☐ the demand is not accompanied by a power of attorney appointing him.

☐ the power of attorney accompanying the demand is not signed by all the applicants for the elected States.

WO 00/34520
PCT/US99/27919

PATENT COOPERATION TREATY

From the INTERNATIONAL BUREAU

PCT

NOTICE INFORMING THE APPLICANT OF THE COMMUNICATION OF THE INTERNATIONAL APPLICATION TO THE DESIGNATED OFFICES

(PCT Rule 47.1(c), first sentence)

To:
GOLDHUSH, Douglas, H.
Nikaido, Marmelstein, Murray & Oram LLP
Metropolitan Square
655 15th Street N.W.
G Street Lobby, Suite 330
Washington, DC 20005-5701
ETATS-UNIS D'AMERIQUE

Date of mailing (day/month/year) 15 June 2000 (15.06.00)	
Applicant's or agent's file reference F8172-9032	**IMPORTANT NOTICE**

International application No.	**International filing date** (day/month/year)	**Priority date** (day/month/year)
PCT/US99/27919	08 December 1999 (08.12.99)	08 December 1998 (08.12.98)

Applicant
UNIVERSITY OF MARYLAND, COLLEGE PARK et al

1. Notice is hereby given that the International Bureau has communicated, as provided in Article 20, the international application to the following designated Offices on the date indicated above as the date of mailing of this Notice:
   AU,CN,JP,KP,KR,US

   In accordance with Rule 47.1(c), third sentence, those Offices will accept the present Notice as conclusive evidence that the communication of the international application has duly taken place on the date of mailing indicated above and no copy of the international application is required to be furnished by the applicant to the designated Office(s).

2. The following designated Offices have waived the requirement for such a communication at this time:
   AE,AL,AM,AP,AT,AZ,BA,BB,BG,BR,BY,CA,CH,CR,CU,CZ,DE,DK,DM,EA,EE,EP,ES,FI,GB,GD,GE,GH,GM,HR,HU,ID,IL,IN,IS,KE,KG,KZ,LC,LK,LR,LS,LT,LU,LV,MA,MD,MG,MK,MN,MW,MX,NO,NZ,OA,PL,PT,RO,RU,SD,SE,SG,SI,SK,SL,TJ,TM,TR,TT,TZ,UA,UG,UZ,VN,YU,ZA,ZW
   The communication will be made to those Offices only upon their request. Furthermore, those Offices do not require the applicant to furnish a copy of the international application (Rule 49.1(a-bis)).

3. Enclosed with this Notice is a copy of the international application as published by the International Bureau on
   15 June 2000 (15.06.00) under No. WO 00/34520

REMINDER REGARDING CHAPTER II (Article 31(2)(a) and Rule 54.2)

If the applicant wishes to postpone entry into the national phase until 30 months (or later in some Offices) from the priority date, **a demand for international preliminary examination** must be filed with the competent International Preliminary Examining Authority before the expiration of 19 months from the priority date.

It is the applicant's sole responsibility to monitor the 19-month time limit.

Note that only an applicant who is a national or resident of a PCT Contracting State which is bound by Chapter II has the right to file a demand for international preliminary examination.

REMINDER REGARDING ENTRY INTO THE NATIONAL PHASE (Article 22 or 39(1))

If the applicant wishes to proceed with the international application in the **national phase**, he must, within 20 months or 30 months, or later in some Offices, perform the acts referred to therein before each designated or elected Office.

For further important information on the time limits and acts to be performed for entering the national phase, see the Annex to Form PCT/IB/301 (Notification of Receipt of Record Copy) and Volume II of the PCT Applicant's Guide.

The International Bureau of WIPO
34, chemin des Colombettes
1211 Geneva 20, Switzerland

Facsimile No. (41-22) 740.14.35

Authorized officer
J. Zahra

Telephone No. (41-22) 338.83.38

PATENT COOPERATION TREATY

From the INTERNATIONAL BUREAU

PCT

NOTIFICATION RELATING TO PRIORITY CLAIM

(PCT Rules 26bis.1 and 26bis.2 and Administrative Instructions, Sections 402 and 409)

To:

GOLDHUSH, Douglas, H.
Nikaido, Marmelstein, Murray & Oram LLP
Metropolitan Square
655 15th Street N.W.
G Street Lobby, Suite 330
Washington, DC 20005-5701
ETATS-UNIS D'AMERIQUE

Date of mailing (day/month/year)	
27 April 2000 (27.04.00)	
Applicant's or agent's file reference	**IMPORTANT NOTIFICATION**
F8172-9032	
International application No.	International filing date (day/month/year)
PCT/US99/27919	08 December 1999 (08.12.99)
Applicant	
UNIVERSITY OF MARYLAND, COLLEGE PARK et al	

The applicant is hereby **notified** of the following in respect of the priority claim(s) made in the international application.

1. [X] **Correction of priority claim.** In accordance with the applicant's notice received on: 05 April 2000 (05.04.00), the following priority claim has been corrected to read as follows:

US 17 December 1998 (17.12.98) 60/112,604

[ ] even though the indication of the number of the earlier application is missing.
[ ] even though the following indication in the priority claim is not the same as the corresponding indication appearing in the priority document:

2. [ ] **Addition of priority claim.** In accordance with the applicant's notice received on: , the following priority claim has been added:

[ ] even though the indication of the number of the earlier application is missing.
[ ] even though the following indication in the priority claim is not the same as the corresponding indication appearing in the priority document:

3. [ ] As a **result of the correction and/or addition** of (a) priority claim(s) under items 1 and/or 2, the (earliest) priority date is:

4. [ ] **Priority claim considered not to have been made.**
[ ] The applicant failed to respond to the Invitation under Rule 26bis.2(a) (Form PCT/IB/316) within the prescribed time limit.
[ ] The applicant's notice was received after the expiration of the prescribed time limit under Rule 26bis.1(a).
[ ] The applicant's notice failed to correct the priority claim so as to comply with the requirements of Rule 4.10.
The applicant may, before the technical preparations for international publication have been completed and subject to the payment of a fee, request the International Bureau to publish, together with the international application, information concerning the priority claim. See Rule 26bis.2(c) and the PCT Applicant's Guide, Volume I, Annex B2(IB).

5. [ ] In case where **multiple priorities** have been claimed, the above item(s) relate to the following priority claim(s):

6. A copy of this notification has been sent to the receiving Office and
[ ] to the International Searching Authority (where the international search report has not yet been issued).
[X] the designated Offices (which have already been notified of the receipt of the record copy).

The International Bureau of WIPO **34, chemin des Colombettes** **1211 Geneva 20, Switzerland**	Authorized officer Beatriz Morariu
Facsimile No. (41-22) 740.14.35	Telephone No. (41-22) 338.83.38

Form PCT/IB/318 (July 1998)

003249737

PATENT COOPERATION TREATY

From the INTERNATIONAL BUREAU

PCT

NOTIFICATION CONCERNING SUBMISSION OR TRANSMITTAL OF PRIORITY DOCUMENT

(PCT Administrative Instructions, Section 411)

To:

GOLDHUSH, Douglas, H.
Nikaido, Marmelstein, Murray & Oram LLP
Metropolitan Square
655 15th Street N.W.
G Street Lobby, Suite 330
Washington, DC 20005-5701
ETATS-UNIS D'AMERIQUE

Date of mailing (day/month/year) 27 April 2000 (27.04.00)	
Applicant's or agent's file reference F8172-9032	**IMPORTANT NOTIFICATION**
International application No. PCT/US99/27919	International filing date (day/month/year) 08 December 1999 (08.12.99)
International publication date (day/month/year) Not yet published	Priority date (day/month/year) 08 December 1998 (08.12.98)
Applicant UNIVERSITY OF MARYLAND, COLLEGE PARK et al	

1. The applicant is hereby notified of the date of receipt (except where the letters "NR" appear in the right-hand column) by the International Bureau of the priority document(s) relating to the earlier application(s) indicated below. Unless otherwise indicated by an asterisk appearing next to a date of receipt, or by the letters "NR", in the right-hand column, the priority document concerned was submitted or transmitted to the International Bureau in compliance with Rule 17.1(a) or (b).

2. This updates and replaces any previously issued notification concerning submission or transmittal of priority documents.

3. An **asterisk(*)** appearing next to a date of receipt, in the right-hand column, denotes a priority document submitted or transmitted to the International Bureau but not in compliance with Rule 17.1(a) or (b). In such a case, **the attention of the applicant is directed** to Rule 17.1(c) which provides that no designated Office may disregard the priority claim concerned before giving the applicant an opportunity, upon entry into the national phase, to furnish the priority document within a time limit which is reasonable under the circumstances.

4. The **letters "NR"** appearing in the right-hand column denote a priority document which was not received by the International Bureau or which the applicant did not request the receiving Office to prepare and transmit to the International Bureau, as provided by Rule 17.1(a) or (b), respectively. In such a case, **the attention of the applicant is directed** to Rule 17.1(c) which provides that no designated Office may disregard the priority claim concerned before giving the applicant an opportunity, upon entry into the national phase, to furnish the priority document within a time limit which is reasonable under the circumstances.

Priority date	Priority application No.	Country or regional Office or PCT receiving Office	Date of receipt of priority document
08 Dece 1998 (08.12.98)	60/111,494	US	10 Marc 2000 (10.03.00)
17 Dece 1998 (17.12.98)	60/112,604	US	10 Marc 2000 (10.03.00)

The International Bureau of WIPO
34, chemin des Colombettes
1211 Geneva 20, Switzerland

Facsimile No. (41-22) 740.14.35

Authorized officer

Beatriz Morariu

Telephone No. (41-22) 338.83.38

PATENT COOPERATION TREATY

From the INTERNATIONAL BUREAU

PCT

INVITATION TO CORRECT
PRIORITY CLAIM

(PCT Rules 4.10, 26*bis*.1, 26*bis*.2(a) and (b))

To:

GOLDHUSH, Douglas, H.
Nikaido, Marmelstein, Murray & Oram LLP
Metropolitan Square
655 15th Street N.W.
G Street Lobby, Suite 330
Washington, DC 20005-5701
ETATS-UNIS D'AMERIQUE

Date of mailing (*day/month/year*) 31 March 2000 (31.03.00)	
Applicant's or agent's file reference F8172-9032	**REPLY DUE** See item 1
International application No. PCT/US99/27919	International filing date (*day/month/year*) 08 December 1999 (08.12.99)
Applicant UNIVERSITY OF MARYLAND, COLLEGE PARK	

The applicant is hereby **invited**, within the time limit indicated below, to correct, by a notice submitted to the International Bureau, defects in the priority claim(s), as indicated in the Annex:

1. **Time limit to respond to this invitation (Rule 26*bis*.1(a)):**
   – within 16 months from the (earliest) priority date; or
   – if the (earliest) priority date is changed as a result of the correction or addition of the (earliest) priority claim, within 16 months from that (earliest) priority date so changed,
   whichever expires first, provided that such a notice may, in any event, be submitted until the expiration of four months from the international filing date.

   Failure to respond to this invitation within the prescribed time limit may result in the priority claim concerned to be considered, for the purposes of the procedure under the PCT, not to have been made (Rule 26*bis*.2(b)).

2. In the case where **multiple priorities** have been claimed, this invitation relates to the following priority claim(s):
   US 15 December 1998 (15.12.98) 60/112,604

3. A copy of this invitation is being sent to the receiving Office.

The International Bureau of WIPO
34, chemin des Colombettes
1211 Geneva 20, Switzerland

Facsimile No. (41-22) 740.14.35

Authorized officer
Athina Nickitas-Etienne

Telephone No. (41-22) 338.83.38

Form PCT/IB/316 (January 2000)

ANNEX TO FORM PCT/IB/316

International application No.
PCT/US99/27919

The International Bureau has found the following defects in the priority claim(s):

1. Failure to Comply with the Requirements of Rule 4.10

a. ☐ **National** application

- ☐ Missing indication of the filing date of the earlier application.
- ☐ Filing date indicated for the earlier application does not fall within the period of 12 months preceding the international filing date.
- ☐ Missing indication of the number of the earlier application.*
- ☐ Missing indication of the country party to the Paris Convention for the Protection of Industrial Property, or of the Member of the World Trade Organization that is not party to that Convention, in which the earlier national application was filed.
- ☐ The country indicated is neither a party to the Paris Convention for the Protection of Industrial Property nor a Member of the World Trade Organization.

b. ☐ **Regional** application

- ☐ Missing indication of the filing date of the earlier application.
- ☐ Filing date indicated for the earlier application does not fall within the period of 12 months preceding the international filing date.
- ☐ Missing indication of the number of the earlier application.*
- ☐ Missing indication of the authority entrusted with the granting of regional patents under the applicable regional patent treaty.
- ☐ The authority indicated as the authority entrusted with the granting of regional patents does not grant regional patents.
- ☐ The priority claim in relation to the ARIPO application does not indicate either at least one country party to the Paris Convention for the Protection of Industrial Property, or at least one Member of the World Trade Organization, for which the earlier application was filed.

c. ☐ **International** application

- ☐ Missing indication of the filing date of the earlier application.
- ☐ Filing date indicated for the earlier application does not fall within the period of 12 months preceding the international filing date.
- ☐ Missing indication of the number of the earlier application.*
- ☐ Missing indication of the receiving Office with which it was filed.

2. Inconsistency with the Corresponding Indications in the Priority Document*

a. ☒ Inconsistency with regard to the filing date of the earlier application:
The request indicates: 15 December 1998 (15. 12.98)
The priority document indicates: 17 December 1998 (17.12.98)

b. ☐ Inconsistency with regard to the number of the earlier application:
The request indicates:
The priority document indicates:

c. ☐ Inconsistency with regard to the country party to the Paris Convention for the Protection of Industrial Property or the Member of the World Trade Organization in which the **national** application was filed:
The request indicates:
The priority document indicates:

d. ☐ Inconsistency with regard to the authority entrusted with the granting of **regional patents** under the applicable regional patent treaty:
The request indicates:
The priority document indicates:

e. ☐ Inconsistency with regard to the receiving Office with which the **international** application was filed:
The request indicates:
The priority document indicates:

* Even if this defect is not corrected in response to this invitation, the priority claim concerned will not be considered not to have been made (Rule 26*bis*.2(b)).

PATENT COOPERATION TREATY

From the INTERNATIONAL BUREAU

PCT

NOTIFICATION CONCERNING SUBMISSION OR TRANSMITTAL OF PRIORITY DOCUMENT

(PCT Administrative Instructions, Section 411)

To:

GOLDHUSH, Douglas, H.
Nikaido, Marmelstein, Murray & Oram LLP
Metropolitan Square
655 15th Street N.W.
G Street Lobby, Suite 330
Washington, DC 20005-5701
ETATS-UNIS D'AMERIQUE

Date of mailing (day/month/year) 23 March 2000 (23.03.00)	
Applicant's or agent's file reference F8172-9032	**IMPORTANT NOTIFICATION**
International application No. PCT/US99/27919	International filing date (day/month/year) 08 December 1999 (08.12.99)
International publication date (day/month/year) Not yet published	Priority date (day/month/year) 08 December 1998 (08.12.98)
Applicant UNIVERSITY OF MARYLAND, COLLEGE PARK et al	

1. The applicant is hereby notified of the date of receipt (except where the letters "NR" appear in the right-hand column) by the International Bureau of the priority document(s) relating to the earlier application(s) indicated below. Unless otherwise indicated by an asterisk appearing next to a date of receipt, or by the letters "NR", in the right-hand column, the priority document concerned was submitted or transmitted to the International Bureau in compliance with Rule 17.1(a) or (b).

2. This updates and replaces any previously issued notification concerning submission or transmittal of priority documents.

3. An **asterisk(*)** appearing next to a date of receipt, in the right-hand column, denotes a priority document submitted or transmitted to the International Bureau but not in compliance with Rule 17.1(a) or (b). In such a case, **the attention of the applicant is directed** to Rule 17.1(c) which provides that no designated Office may disregard the priority claim concerned before giving the applicant an opportunity, upon entry into the national phase, to furnish the priority document within a time limit which is reasonable under the circumstances.

4. The **letters "NR"** appearing in the right-hand column denote a priority document which was not received by the International Bureau or which the applicant did not request the receiving Office to prepare and transmit to the International Bureau, as provided by Rule 17.1(a) or (b), respectively. In such a case, **the attention of the applicant is directed** to Rule 17.1(c) which provides that no designated Office may disregard the priority claim concerned before giving the applicant an opportunity, upon entry into the national phase, to furnish the priority document within a time limit which is reasonable under the circumstances.

Priority date	Priority application No.	Country or regional Office or PCT receiving Office	Date of receipt of priority document
08 Dece 1998 (08.12.98)	60/111,494	US	10 Marc 2000 (10.03.00)

The International Bureau of WIPO **34, chemin des Colombettes** **1211 Geneva 20, Switzerland**	Authorized officer Tessadel PAMPLIEGA
Facsimile No. (41-22) 740.14.35	Telephone No. (41-22) 338.83.38

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US99/27919

A. CLASSIFICATION OF SUBJECT MATTER

IPC(7) : C12Q 1/68; C07H 21/04
US CL : 435/6; 536/23.5

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
U.S. : 435/6; 536/23.5

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)
Please See Continuation Sheet

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	BJORBAEK et al. Genetic Variants in Promoters and Coding Regions of the Muscle Glycogen Synthase and the Insulin-Responsive GLUT4 Genes in NIDDM. Diabetes. August 1994, Vol. 43, pages 976-983.	1
A	OSSEI-GERNING et al. Insulin receptor substrate-1 gene polymorphism and cardiovascular risk in non-insulin dependent diabetes mellitus and patients undergoing coronary angiography. Clin. Lab. Haem. 1997, Vol. 19, pages 123-129.	3
A	GROBET et al. Molecular definition of an allelic series of mutations disrupting the myostatin function and causing double-muscling in cattle. Mammalian Genome. 1998, Vol. 9, pages 210-213.	2, 4

☐ Further documents are listed in the continuation of Box C. ☐ See patent family annex.

* Special categories of cited documents:

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier application or patent published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search
08 March 2000 (08.03.2000)

Date of mailing of the international search report
05 APR 2000

Name and mailing address of the ISA/US
Commissioner of Patents and Trademarks
Box PCT
Washington, D.C. 20231
Facsimile No. (703) 305-3230

Authorized officer
Juliet C. Einsmann
Telephone No. (703) 308-0196

Form PCT/ISA/210 (second sheet) (July 1998)

PATENT COOPERATION TREATY

09/856470 **PCT**

15

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Article 36 and Rule 70)

Applicant's or agent's file reference 108172-09032	**FOR FURTHER ACTION**	See Notification of Transmittal of International Preliminary Examination Report (Form PCT/IPEA/416)
International application No. PCT/US99/27919	International filing date *(day/month/year)* 08 December 1999 (08.12.1999)	Priority date *(day/month/year)* 08 December 1998 (08.12.1998)
International Patent Classification (IPC) or national classification and IPC IPC(7): C12Q 1/68; C07H 21/04 and US Cl.: 435/6; 536/23.5		
Applicant UNIVERSITY OF MARYLAND, COLLEGE PARK		

1. This international preliminary examination report has been prepared by this International Preliminary Examining Authority and is transmitted to the applicant according to Article 36.

2. This REPORT consists of a total of 3 sheets, including this cover sheet.

   ☐ This report is also accompanied by ANNEXES, i.e., sheets of the description, claims and/or drawings which have been amended and are the basis for this report and/or sheets containing rectifications made before this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions under the PCT).

   These annexes consist of a total of 9 sheets.

3. This report contains indications relating to the following items:

   - I ☒ Basis of the report
   - II ☐ Priority
   - III ☐ Non-establishment of report with regard to novelty, inventive step and industrial applicability
   - IV ☐ Lack of unity of invention
   - V ☒ Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
   - VI ☐ Certain documents cited
   - VII ☐ Certain defects in the international application
   - VIII ☐ Certain observations on the international application

Date of submission of the demand	Date of completion of this report
07 July 2000 (07.07.2000)	24 November 2000 (24.11.2000)
Name and mailing address of the IPEA/US Commissioner of Patents and Trademarks Box PCT Washington, D.C. 20231 Facsimile No. (703)305-3230	Authorized officer Juliet C. Einsmann Telephone No. (703) 308-01

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

International application No.
PCT/US99/27919

I. Basis of the report

1. With regard to the **elements** of the international application:*

☒ the international application as originally filed.

☒ the description:
pages 1-10 as originally filed
pages NONE , filed with the demand
pages NONE , filed with the letter of _____.

☒ the claims:
pages 11 and 12 , as originally filed
pages NONE , as amended (together with any statement) under Article 19
pages NONE , filed with the demand
pages NONE , filed with the letter of _____.

☒ the drawings:
pages NONE , as originally filed
pages NONE , filed with the demand
pages NONE , filed with the letter of _____.

☐ the sequence listing part of the description:
pages NONE , as originally filed
pages NONE , filed with the demand
pages NONE , filed with the letter of _____.

2. With regard to the **language**, all the elements marked above were available or furnished to this Authority in the language in which the international application was filed, unless otherwise indicated under this item.
These elements were available or furnished to this Authority in the following language _____ which is:

☐ the language of a translation furnished for the purposes of international search (under Rule23.1(b)).

☐ the language of publication of the international application (under Rule 48.3(b)).

☐ the language of the translation furnished for the purposes of international preliminary examination(under Rules 55.2 and/or 55.3).

3. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, the international preliminary examination was carried out on the basis of the sequence listing:

☐ contained in the international application in printed form.

☐ filed together with the international application in computer readable form.

☐ furnished subsequently to this Authority in written form.

☐ furnished subsequently to this Authority in computer readable form.

☐ The statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.

☐ The statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished.

4. ☐ The amendments have resulted in the cancellation of:

☐ the description, pages NONE

☐ the claims, Nos. NONE

☐ the drawings, sheets/~~fig~~ NONE

5. ☐ This report has been established as if (some of) the amendments had not been made, since they have been considered to go beyond the disclosure as filed, as indicated in the Supplemental Box (Rule 70.2(c)).**

** Replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report since they do not contain amendments (Rules 70.16 and 70.17).*
*** Any replacement sheet containing such amendments must be referred to under item 1 and annexed to this report.*

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

International application No.
PCT/US99/27919

V. Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. STATEMENT

Novelty (N)	Claims 1-4	YES
	Claims NONE	NO
Inventive Step (IS)	Claims 1-4	YES
	Claims NONE	NO
Industrial Applicability (IA)	Claims 1-4	YES
	Claims NONE	NO

2. CITATIONS AND EXPLANATIONS (Rule 70.7)

Claims 1-4 meet the criteria set out in PCT Articles 33(2)-(3) since the prior art does not teach or fairly suggest the claimed invention. The cited references teach mutations in the glucose transport 4 gene (Bjorbaek et al.), the myostatin exon 2 gene (Grobet et al.), and the insulin receptor substrate-1 gene (Ossei-Gerning et al.). The prior art fails to teach, however, an association between these mutations and high cholesterol or that people with specific genotypes in these genes will be more responsive to specific exercise levels for improving choleterol levels.

Claims 1-4 meet the criteria set out in PCT Article 33(4) for industrial applicability.

---------- NEW CITATIONS ----------

NONE

PCT WORLD INTELLECTUAL PROPERTY ORGANIZATION
International Bureau

INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) **International Patent Classification** 7 :
C12Q 1/68, C07H 21/04

A1

(11) **International Publication Number:** **WO 00/34520**

(43) **International Publication Date:** 15 June 2000 (15.06.00)

(21) **International Application Number:** PCT/US99/27919

(22) **International Filing Date:** 8 December 1999 (08.12.99)

(30) **Priority Data:**

60/111,494	8 December 1998 (08.12.98)	US
60/112,604	17 December 1998 (17.12.98)	US

(71) **Applicants** *(for all designated States except US)*: UNIVERSITY OF MARYLAND, COLLEGE PARK [US/US]; Office of Technology Liaison, 4312 Knox Road, College Park, MD 20742 (US). UNIVERSITY OF MARYLAND, BALTIMORE [US/US]; Office of Research & Development, 515 West Lombard Street, Baltimore, MD 21201 (US). UNIVERSITY OF PITTSBURGH [US/US]; Office of Technology Management, Rm 9110, William Pitt Union, Pittsburgh, PA 15260 (US).

(72) **Inventors; and**
(75) **Inventors/Applicants** *(for US only)*: HAGBERG, James, M. [US/US]; 10941 Hilltop Lane, Columbia, MD 21044 (US). FERRELL, Robert, E. [US/US]; 206 Maple Avenue, Pittsburgh, PA 15218 (US). SHULDINER, Alan [US/US]; 10600 Harpoon Hill, Columbia, MD 21044 (US).

(74) **Agents:** GOLDHUSH, Douglas, H. et al.; Nikaido, Marmelstein, Murray & Oram LLP, Metropolitan Square, 655 15th Street N.W., G Street Lobby, Suite 330, Washington, DC 20005–5701 (US).

(81) **Designated States:** AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CR, CU, CZ, DE, DK, DM, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, ARIPO patent (GH, GM, KE, LS, MW, SD, SL, SZ, TZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG).

Published
With international search report.
Before the expiration of the time limit for amending the claims and to be republished in the event of the receipt of amendments.

(54) **Title:** GENETIC MARKERS WHICH IDENTIFY INDIVIDUALS WHO IMPROVE THEIR CHOLESTEROL LEVELS AND DIABETES STATUS WITH EXERCISE

(57) **Abstract**

A method of improving cholesterol levels or diabetes status in subjects with hypercholesteremia or diabetes or at risk of developing such disorders includes identifying subjects having an allele and/or a genotype at a gene locus which positively correlates with greater success in improving cholesterol levels or diabetes status in hypercholesteremic or diabetic individuals, respectively, as compared with other alleles and/or genotypes at the same gene locus, and engaging the subject in exercise training for a period of time sufficient to improve the subject's cholesterol levels or diabetes status. Genotypic distinctions may be found, for example, in the glucose transport 4 gene, the myostatin exon 2 gene and the insulin receptor substrate–1 (IRS–1) gene.

FOR THE PURPOSES OF INFORMATION ONLY

Codes used to identify States party to the PCT on the front pages of pamphlets publishing international applications under the PCT.

AL	Albania	**ES**	Spain	**LS**	Lesotho	**SI**	Slovenia
AM	Armenia	**FI**	Finland	**LT**	Lithuania	**SK**	Slovakia
AT	Austria	**FR**	France	**LU**	Luxembourg	**SN**	Senegal
AU	Australia	**GA**	Gabon	**LV**	Latvia	**SZ**	Swaziland
AZ	Azerbaijan	**GB**	United Kingdom	**MC**	Monaco	**TD**	Chad
BA	Bosnia and Herzegovina	**GE**	Georgia	**MD**	Republic of Moldova	**TG**	Togo
BB	Barbados	**GH**	Ghana	**MG**	Madagascar	**TJ**	Tajikistan
BE	Belgium	**GN**	Guinea	**MK**	The former Yugoslav Republic of Macedonia	**TM**	Turkmenistan
BF	Burkina Faso	**GR**	Greece			**TR**	Turkey
BG	Bulgaria	**HU**	Hungary	**ML**	Mali	**TT**	Trinidad and Tobago
BJ	Benin	**IE**	Ireland	**MN**	Mongolia	**UA**	Ukraine
BR	Brazil	**IL**	Israel	**MR**	Mauritania	**UG**	Uganda
BY	Belarus	**IS**	Iceland	**MW**	Malawi	**US**	United States of America
CA	Canada	**IT**	Italy	**MX**	Mexico	**UZ**	Uzbekistan
CF	Central African Republic	**JP**	Japan	**NE**	Niger	**VN**	Viet Nam
CG	Congo	**KE**	Kenya	**NL**	Netherlands	**YU**	Yugoslavia
CH	Switzerland	**KG**	Kyrgyzstan	**NO**	Norway	**ZW**	Zimbabwe
CI	Côte d'Ivoire	**KP**	Democratic People's Republic of Korea	**NZ**	New Zealand		
CM	Cameroon			**PL**	Poland		
CN	China	**KR**	Republic of Korea	**PT**	Portugal		
CU	Cuba	**KZ**	Kazakstan	**RO**	Romania		
CZ	Czech Republic	**LC**	Saint Lucia	**RU**	Russian Federation		
DE	Germany	**LI**	Liechtenstein	**SD**	Sudan		
DK	Denmark	**LK**	Sri Lanka	**SE**	Sweden		
EE	Estonia	**LR**	Liberia	**SG**	Singapore		

09/856470
JC18 Rec'd PCT/PTO 07 JUN 2001
PCT/US99/27919

GENETIC MARKERS WHICH IDENTIFY INDIVIDUALS WHO IMPROVE THEIR CHOLESTEROL LEVELS AND DIABETES STATUS WITH EXERCISE

FIELD OF THE INVENTION

The present invention relates to identifying one or more genetic markers which correlate with greater success in improving cholesterol levels and diabetes status in individuals with and without high cholesterol levels or diabetes.

BACKGROUND OF THE INVENTION

Studies have shown that individuals suffering from or at risk of developing high cholesterol levels or diabetes can alleviate symptoms or otherwise improve their conditions through exercise. Unfortunately, some individuals, no matter how rigorously they exercise, are unable to improve their conditions, while others benefit to a much greater extent than predicted. These results underscore the fact that many factors contribute to an individual's well-being. Such factors include, for example, behaviors such as diet and exercise, genetic makeup, and environment. While behavior and environment can be controlled, altered or regulated, an individual's genetic makeup is essentially predetermined and set at birth. The present inventors hypothesized that upon identifying the genetic makeup of a population suffering from or at risk of developing high cholesterol levels or diabetes and observing that some individuals of the population improve their cholesterol levels and diabetic status from a change of behavior to a much greater or lesser extent than expected, a correlation could be made between the presence or absence of certain genetic markers and success in improving cholesterol levels and diabetic status.

An object of the present invention is to identify one or more genetic markers which positively correlate with greater success in improving cholesterol levels and diabetes status in individuals with and without high cholesterol levels or diabetes.

SUMMARY OF THE INVENTION

The present inventors have discovered a number of genetic markers which positively correlate with greater success in improving cholesterol levels and diabetes status in diabetic, hypercholesteremic or at risk individuals, as compared with other genetic makeup at the same gene loci. Therefore, a first embodiment of the present invention is directed to a method of improving cholesterol levels in a subject with increased cholesterol levels or at risk of developing such a condition, the method comprising:

identifying a subject with hypercholesteremia or at risk of developing such a condition having an allele and/or a genotype at a gene locus which positively correlates with greater success in improving cholesterol levels in hypercholesteremic individuals, as compared with other alleles and/or genotypes at the same gene locus; and

engaging the subject in exercise training for a period of time sufficient to improve the cholesterol levels in the subject.

A second embodiment of the present invention is directed to a method of improving diabetes status in a subject with diabetes or at risk of developing diabetes, the method comprising:

identifying a subject with diabetes or at risk of developing diabetes having an allele and/or a genotype at a gene locus which positively correlates with greater success in improving diabetes status in diabetic individuals, as compared with other alleles and/or genotypes at the same gene locus; and

engaging the subject in exercise training for a period of time sufficient to improve the diabetes status in the subject.

DETAILED DESCRIPTION OF THE INVENTION

The inventors have found that a number of genetic markers positively correlate with greater success in improving cholesterol levels or diabetes status in individuals with hypercholesteremia or diabetes, or at risk of developing such disorders, as compared with other genetic makeup at the same gene loci.

Markers which the inventors have investigated include the glucose transport 4 (GLUT4) gene, the myostatin gene and the insulin receptor substrate-1 (IRS-1) gene.

The term "improved cholesterol levels" means an improvement in at least one characteristic area which is associated with hypercholesteremia. An improvement may be in one or more of the following characteristic areas (this list is non-exhaustive and includes overlapping and representative examples only): change in cholesterol metabolism, increase in high density lipoprotein cholesterol (HDL-C) levels, increase in high density lipoprotein cholesterol 2 (HDL_2-C) levels, decrease in low density lipoprotein cholesterol (LDL-C) levels or increase in the ratio of HDL-C or HDL_2-C levels as compared to LDL-C levels. These improvements may be measured by, for example, plasma cholesterol tests conducted before and after exercise training. An improvement in cholesterol levels in accordance with the invention may be found both in individuals with hypercholesteremia and in individuals at risk of developing such a disorder.

The term "improved diabetes status" means an improvement in at least one characteristic area which is associated with diabetes. An improvement may be in one or more of the following characteristic areas (this list is non-exhaustive and includes overlapping and representative examples only): change in glucose metabolism, change in insulin metabolism, change in glucose levels from a baseline determination, change in insulin levels from a baseline determination, change in fasting plasma glucose levels, change in fasting plasma insulin levels or change in acute insulin response to glucose. These improvements may be measured by, for example, glucose tolerance tests conducted before and after exercise training. An improvement in diabetes status in accordance with the invention may be found both in individuals with diabetes and in individuals at risk of developing diabetes.

The term "single course of exercise", as used throughout this application, means a cardiovascular exercise session of any type which is conducted during

one day. An exercise session may comprise an aerobics class, treadmill training, step machine, or any other suitable cardiovascular exercise regimen. For most cases, exercise may be completed in, for example, 30 minutes to 3 hours, with optional brief rest periods of 3-15 minutes, however this amount would vary depending on the health and endurance of the subject.

The term "extensive exercise" means about 10 single courses of exercise or more, preferably at least 15, at least 20, or at least 25 single courses of exercise, over a defined period of time ("the exercise period"). The exercise period in the case of an extensive exercise protocol may be from about 50-400 days, preferably from about 70-350 days or 100-300 days.

The time between exercise courses depends on the health and endurance of the subject. Preferably, the time between exercise courses may be from about 1-3 days or more.

The present inventors have discovered that hypercholesteremic or diabetic individuals or those at risk of developing hypercholesteremia or diabetes with different genotypes for genes which control the manufacture of certain proteins exhibit different degrees of success in improving their cholesterol levels and diabetes status through exercise. The inventors have surprisingly discovered that each genotype potentially can benefit from exercise, however, the amount of exercise which produces the most benefits varies according to genotype. These results could not have been predicted from initial patient screening.

Glucose transport in skeletal muscle is mainly facilitated by the insulin-responsive GLUT4. In the basal state, GLUT4 is stored in a transporter-enriched intracellular pool. Following insulin stimulation, GLUT4 is translocated from the intracellular compartment to both the plasma membrane and the T-tubules.

The inventors have found that subjects having a BamHI "AA" genotype for a GLUT4 gene exhibit a greater improvement in cholesterol levels than those with a "GG" or "AG" genotype, after extensive exercise.

Therefore, one method of improving cholesterol levels in a subject in need of such improvement according to the invention comprises identifying a subject having a BamHI "AA" genotype for a GLUT4 gene, wherein the subject is in need of improved cholesterol levels and engaging the subject in extensive exercise training for a period of time sufficient to improve the cholesterol levels in the subject.

Myostatin, also known as growth/differentiation factor-8 (Gdf8), is a member of the transforming growth factor-beta (TGF-β) superfamily, which encompasses a large number of growth and differentiation factors that play important roles in regulating embryonic development and in maintaining tissue homeostasis in adult animals. During early stages of embryogenesis, myostatin expression is restricted to the myotome compartment of developing somites. At later stages and in adult animals, myostatin is expressed in many different muscles throughout the body.

The inventors have found that subjects having a "12" genotype for exon 2 of the myostatin gene exhibit a greater improvement in cholesterol levels than those with a "11" genotype, after extensive exercise.

Therefore, another method of improving cholesterol levels in a subject in need of such improvement according to the invention comprises identifying a subject having a "12" genotype for exon 2 of a myostatin gene, wherein the subject is in need of improved cholesterol levels and engaging the subject in extensive exercise training for a period of time sufficient to improve the cholesterol levels in the subject.

The inventors have also found that diabetics or those at risk of developing diabetes having a "11" genotype at exon 2 of the myostatin gene improve their diabetes status more with extensive exercise training than those having a "12" genotype.

Therefore, a method of improving diabetes status in a subject in need of such improvement comprises identifying a subject having a "11" genotype for exon 2 of a myostatin gene, wherein the subject is in need of improved diabetes

status and engaging the subject in extensive exercise training for a period of time sufficient to improve the diabetes status in the subject.

IRS-1 is a 185 kDa protein which is activated rapidly upon insulin stimulation of cells, and is a key mediator of an insulin-regulated biological activity. The amino-terminal region of the protein contains interaction modules that facilitate its binding to receptors of insulin. The remainder of the molecule contains numerous tyrosine containing motifs, which, when phosphorylated by the insulin receptor tyrosine kinase, serve as binding regions for a variety of cellular proteins containing a so-called "SH2" domain.

The inventors have found that subjects having a "12" genotype for the IRS-1 gene exhibit a greater improvement in cholesterol levels than those with a "11" genotype, after extensive exercise.

Therefore, in accordance with this aspect of the present invention, a method of improving cholesterol levels in a subject in need of such improvement comprises identifying a subject having a "12" genotype for an IRS-1 gene, wherein the subject is in need of improved cholesterol levels and engaging the subject in extensive exercise training for a period of time sufficient to improve the cholesterol levels in the subject.

EXAMPLES

Example 1. Variations in Improvement of Cholesterol Levels in Subjects with Different GLUT4 BamHI, Myostatin and IRS-1 Genotypes After Extensive Exercise

DNA was obtained from obese sedentary men 50-65 yrs of age, and processed as follows.

Detection of (C581T) and (A30G) Substitution in GLUT4

Genotyping for the (C581T) and (A30G) substitutions in GLUT4 was performed by amplification using sense primer 5'-CAGTGCCCGGAGCAGGGAGGCGCT-3' and antisense primer 5'-

GCGAAGATGAAAGAACCGATCCTG-3' followed by digestion with the restriction endonucleases AvaII and BamHI, respectively. The presence of a C at np-518 yields major AvaII restriction fragments of 408 and 378 base pairs and the presence of a T at np-518 yields fragments of 119, 289 and 378 base pairs. The presence of an A at np-30 yields major BamHI fragments of 389 and 342 base pairs, while the presence of a G at np-30 yields fragments of 445 and 389 base pairs. All denotations of sequence positions are based on those recited in Bjorback et al. (1994), *Diabetes*, 43:976-983, hereby incorporated by reference.

Detection of Lys153Arg Substitution in Myostatin Exon 2

DNA amplification primers for exon 2 of the human myostatin gene were designed based on the cDNA sequence of human myostatin (GenBank Accession No. AF019627) and the genomic organization of the bovine myostatin gene (Grobert et al. (1998), *Mamm. Gen.* 9:210-213, incorportated by reference). Amplimers were sequenced directly using the dRhodamine ready reaction kit (Perkin Elmer) and analyzed on the ABI Prism Model 377 (Applied Biosystems) fluorescent sequencer. Sequences were aligned for comparison using SEQUENCHER™ 3.0 (Gene Codes). Primer 1 had the sequence 5'-GAAAACCCAAATGTTGCTTC-3', and primer 2 had the sequence 5'-TGTCTAGCTTATGAGCTTAGGG-3'. The temperature was 54°C, and the buffer was 2.0 mM MgCl. PCR products were digested with BanII and the digested products were run on 2% agarose gels.

Detection of Gly972Arg Substitution in IRS-1

A 220 bp region encompassing the Gly972Arg substitution was amplified from approximately 20 ng of genomic DNA with upstream primer 5'-GCAGCCTGGCAGGAGAGCCAT-3' and downstream primer 5'-CTCACCTCCTCTGCAGCAATG-3'. PCR products were digested with BstNI. The digested products were run on a 4% agarose gel, stained with

ethidium bromide, and visualized by UV transillumination. The expected digestion product sizes were 220 bp for Gly972 homozygotes, 164 bp and 56 bp for Arg972 homozygotes and 220 bp, 164 bp and 56 bp for heterozygotes.

Results

The subjects underwent 9 months of endurance exercise training to quantify, among other things, their improvements in plasma cholesterol levels. Subjects were initially stabilized on an American Heart Association low-fat diet and had fasting blood samples drawn for plasma cholesterol measurements. This diet was maintained throughout the 9 months of exercise training and subjects repeated the blood sampling for cholesterol levels after training. The data in Table 1 represent the change in HDL-C and HDL_2-C levels that occurred with exercise training. Subjects with the GLUT4 BamHI "AA" genotype increased their plasma HDL-C and HDL_2-C levels with exercise training substantially more than subjects with the GLUT4 BamHI "GG" or "AG" genotype. Furthermore, subjects with the myostatin exon 2 "12" genotype increased their plasma HDL-C and HDL_2-C levels with exercise training substantially more than subjects with the myostatin exon 2 "11" genotype. Lastly, subjects with the IRS-1 "12" genotype increased their plasma HDL-C and HDL_2-C levels with exercise training substantially more than subjects with the IRS-1 "11" genotype. Thus, these results indicate that GLUT4, myostatin exon 2 and IRS-1 genotypes identify those individuals most likely to improve their cholesterol levels with exercise training.

Table 1: Change with Exercise Training in Plasma Lipoprotein Levels as a Function of Genotype

	Change with Exercise Training	
GLUT4 BamHI	**HDL-C**	**HDL_2-C**

GG and AG genotype (n=13)	1.9 ± 3.8	0.7 ± 4.0
AA genotype (n=2)	16.9 ± 12.9	11.1 ± 15.0
Myostatin Exon 2		
11 genotype (n=13)	2.0 ± 4.0	0.4 ± 4.0
12 genotype (n=2)	15.8 ± 14.4	13.6 ± 11.5
IRS-1		
11 genotype (n=10)	2.9 ± 1.4	-0.1 ± 1.5
12 genotype (n=3)	11.4 ± 7.3	8.5 ± 6.6

Values are mean ± SD. Values are expressed as the change with 9 months of exercise training in HDL-C and HDL_2-C levels. Thus, positive values indicate a response that is greater after training.

Example 2. Variations in Improvement of Diabetes Status in Subjects with Different Myostatin Exon 2 Genotypes After Extensive Exercise

The subjects, detection of polymorphisms and the exercise regimen for these subjects was described in Example 1. Subjects underwent an oral glucose tolerance test with blood samples drawn for up to 3 hours after the ingestion of a standard glucose load. Subjects repeated the glucose tolerance test after training. The data in the following Table 2 represent the change in the integrated glucose area above baseline that occurred with the exercise training. Subjects with the myostatin exon 2 "11" genotype decreased their glucose areas more with exercise training than subjects with the myostatin exon 2 "12" genotype. These results indicate that myostatin exon 2 genotypes identify those individuals most likely to improve their diabetes status with exercise training.

Table 2: Change with Exercise Training in Integrated Glucose Area in Response to an Oral Glucose Tolerance Test as a Function of Genotype

Myostatin Exon 2 Genotype	Change with Exercise Training in Glucose Area
11 genotype (n=14)	-1941 ± 1260
12 genotype (n=3)	1180 ± 1641

Values are mean ± SD. Values are expressed as the change with 9 months of exercise training in integrated glucose area above baseline for 3 hours following a standard oral glucose challenge. Negative values indicate a response that is reduced after exercise training and positive values a response that is greater after training.

We claim:

1. A method of improving cholesterol levels in a subject in need of such improvement, the method comprising:

identifying a subject with hypercholesteremia or at risk of developing hypercholesteremia having a BamHI "AA" genotype for a glucose transport 4 gene, wherein the subject is in need of improved cholesterol levels; and

engaging the subject in extensive exercise training for a period of time sufficient to improve the cholesterol levels in the subject.

2. A method of improving cholesterol levels in a subject in need of such improvement, the method comprising:

identifying a subject with hypercholesteremia or at risk of developing hypercholesteremia having a "12" genotype for a myostatin exon 2 gene, wherein the subject is in need of improved cholesterol levels; and

engaging the subject in extensive exercise training for a period of time sufficient to improve the cholesterol levels in the subject.

3. A method of improving cholesterol levels in a subject in need of such improvement, the method comprising:

identifying a subject with hypercholesteremia or at risk of developing hypercholesteremia having a "12" genotype for an insulin receptor substrate-1 gene, wherein the subject is in need of improved cholesterol levels; and

engaging the subject in extensive exercise training for a period of time sufficient to improve the cholesterol levels in the subject.

4. A method of improving diabetes status in a subject in need of such improvement, the method comprising:

identifying a subject with diabetes or at risk of developing diabetes having a "11" genotype for a myostatin exon 2 gene, wherein the subject is in need of improved diabetes status; and

engaging the subject in extensive exercise training for a period of time sufficient to improve the diabetes status in the subject.

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US99/27919

A. CLASSIFICATION OF SUBJECT MATTER

IPC(7) : C12Q 1/68; C07H 21/04
US CL : 435/6; 536/23.5

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
U.S. : 435/6; 536/23.5

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)
Please See Continuation Sheet

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	BJORBAEK et al. Genetic Variants in Promoters and Coding Regions of the Muscle Glycogen Synthase and the Insulin-Responsive GLUT4 Genes in NIDDM. Diabetes. August 1994, Vol. 43, pages 976-983.	1
A	OSSEI-GERNING et al. Insulin receptor substrate-1 gene polymorphism and cardiovascular risk in non-insulin dependent diabetes mellitus and patients undergoing coronary angiography. Clin. Lab. Haem. 1997, Vol. 19, pages 123-129.	3
A	GROBET et al. Molecular definition of an allelic series of mutations disrupting the myostatin function and causing double-muscling in cattle. Mammalian Genome. 1998, Vol. 9, pages 210-213.	2, 4

☐ Further documents are listed in the continuation of Box C. ☐ See patent family annex.

* Special categories of cited documents:

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier application or patent published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
08 March 2000 (08.03.2000)	05 APR 2000
Name and mailing address of the ISA/US Commissioner of Patents and Trademarks Box PCT Washington, D.C. 20231 Facsimile No. (703) 305-3230	Authorized officer Juliet C. Einsmann Telephone No. (703) 308-0196

Form PCT/ISA/210 (second sheet) (July 1998)

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US99/27919

Continuation of B. FIELDS SEARCHED Item 3: MEDLINE, BIOSIS, EMBASE, CAPLUS, SCISEARCH
search terms: cholesterol, exercise, diabetes, hypercholestermia, glucose transport, myostatin, insulin receptor substrate

PATENT COOPERATION TREATY

PCT

From the INTERNATIONAL BUREAU

To:

GOLDHUSH, Douglas, H.
Arent Fox Kintner Plotkin & Kahn,
PLLC
1050 Connecticut Avenue, NW
Suite 600
Washington, DC 20036-5339
ETATS-UNIS D'AMERIQUE

NOTIFICATION OF THE RECORDING OF A CHANGE

(PCT Rule 92bis.1 and
Administrative Instructions, Section 422)

Date of mailing (day/month/year)
25 September 2000 (25.09.00)

Applicant's or agent's file reference
F8172-9032

IMPORTANT NOTIFICATION

International application No.
PCT/US99/27919

International filing date (day/month/year)
08 December 1999 (08.12.99)

1. The following indications appeared on record concerning:

[ ] the applicant [ ] the inventor [X] the agent [ ] the common representative

Name and Address
GOLDHUSH, Douglas, H.
Arent Fox Kitner Plotkin & Kahn,
PLLC
1050 Connecticut Avenue, NW
Suite 600
Washington, DC 20005-5701
United States of America

State of Nationality:
State of Residence:
Telephone No.: 202 638 5000
Facsimile No.: 202 638 4810
Teleprinter No.:

2. The International Bureau hereby notifies the applicant that the following change has been recorded concerning:

[ ] the person [ ] the name [X] the address [ ] the nationality [ ] the residence

Name and Address
GOLDHUSH, Douglas, H.
Arent Fox Kintner Plotkin & Kahn,
PLLC
1050 Connecticut Avenue, NW
Suite 600
Washington, DC 20036-5339
United States of America

State of Nationality:
State of Residence:
Telephone No.: 202 857 6000
Facsimile No.: 202 638 4810
Teleprinter No.:

3. Further observations, if necessary:

4. A copy of this notification has been sent to:

[X] the receiving Office
[ ] the International Searching Authority
[X] the International Preliminary Examining Authority
[ ] the designated Offices concerned
[X] the elected Offices concerned
[ ] other:

The International Bureau of WIPO
34, chemin des Colombettes
1211 Geneva 20, Switzerland
Facsimile No.: (41-22) 740.14.35

Authorized officer
Jocelyne Rey-Millet
Telephone No.: (41-22) 338.83.38

PATENT COOPERATION TREATY

PCT

From the INTERNATIONAL BUREAU

To:

GOLDHUSH, Douglas, H.
Arent Fox Kitner Plotkin & Kahn, PLLC
1050 Connecticut Avenue, NW
Suite 600
Washington, DC 20005-5701
ETATS-UNIS D'AMERIQUE

INFORMATION CONCERNING ELECTED OFFICES NOTIFIED OF THEIR ELECTION

(PCT Rule 61.3)

Date of mailing (day/month/year) 24 August 2000 (24.08.00)	
Applicant's or agent's file reference F8172-9032	**IMPORTANT INFORMATION**

International application No.	**International filing date** (day/month/year)	**Priority date** (day/month/year)
PCT/US99/27919	08 December 1999 (08.12.99)	08 December 1998 (08.12.98)

Applicant
UNIVERSITY OF MARYLAND, COLLEGE PARK et al

1. The applicant is hereby informed that the International Bureau has, according to Article 31(7), notified each of the following Offices of its election:

   AP :GH,GM,KE,LS,MW,SD,SL,SZ,TZ,UG,ZW
   EP :AT,BE,CH,CY,DE,DK,ES,FI,FR,GB,GR,IE,IT,LU,MC,NL,PT,SE
   National :AU,BG,BR,CA,CN,CZ,DE,IL,JP,KP,KR,MN,NO,NZ,PL,RO,RU,SE,SK,US

2. The following Offices have waived the requirement for the notification of their election; the notification will be sent to them by the International Bureau *only upon their request*:

   EA :AM,AZ,BY,KG,KZ,MD,RU,TJ,TM
   OA :BF,BJ,CF,CG,CI,CM,GA,GN,GW,ML,MR,NE,SN,TD,TG
   National :AE,AL,AM,AT,AZ,BA,BB,BY,CH,CR,CU,DK,DM,EE,ES,FI,GB,GD,GE,GH,GM, HR,HU,ID,IN,IS,KE,KG,KZ,LC,LK,LR,LS,LT,LU,LV,MA,MD,MG,MK,MW,MX,PT,SD,SG, SI,SL,TJ,TM,TR,TT,TZ,UA,UG,UZ,VN,YU,ZA,ZW

3. The applicant is reminded that he must enter the "national phase" **before the expiration of 30 months from the priority date** before each of the Offices listed above. This must be done by paying the national fee(s) and furnishing , if prescribed, a translation of the international application (Article 39(1)(a)), as well as, where applicable, by furnishing a translation of any annexes of the international preliminary examination report (Article 36(3)(b) and Rule 74.1).

   Some offices have fixed time limits expiring later than the above-mentioned time limit. For detailed information about the applicable time limits and the acts to be performed upon entry into the national phase before a particular Office, see Volume II of the PCT Applicant's Guide.

   The entry into the European regional phase is postponed **until 31 months from the priority date** for all States designated for the purposes of obtaining a European patent.

The International Bureau of WIPO
34, chemin des Colombettes
1211 Geneva 20, Switzerland

Facsimile No. (41-22) 740.14.35

Authorized officer:
Manu Berrod

Telephone No. (41-22) 338.83.38